Pergamon

Bioorganic & Medicinal Chemistry Letters 11 (2001) 2017–2019

BIOORGANIC & MEDICINAL CHEMISTRY LETTERS

# Substituted 4-Phenyl-2-(phenylcarboxamido)-1,3-thiazole Derivatives as Antagonists for the Adenosine $A_1$ Receptor

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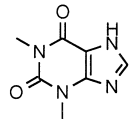
Received 21 March 2001; revised 4 May 2001; accepted 17 May 2001

**Abstract**—The synthesis and receptor binding of novel adenosine receptor antagonists is described. We found that non-xanthine 4-phenyl-2-(phenylcarboxamido)-1,3-thiazole derivatives may have high affinity and substantial selectivity for the adenosine $A_1$ receptor. 

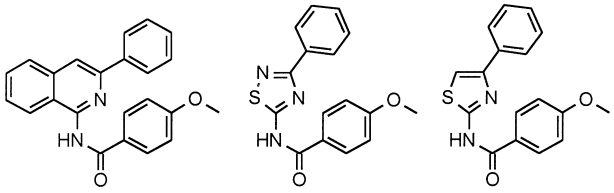
Extracellular adenosine regulates several physiological functions by activation of specific cell membrane receptors. There are four adenosine receptor subclasses defined, $A_1$, $A_{2A}$, $A_{2B}$ and $A_3$. Since the first reports on the adenosine $A_1$ receptor,[1,2] efforts have been made to identify ligands for this receptor. The first $A_1$ receptor antagonists were xanthine derivatives, such as theophylline (**1**, Fig. 1). Since then, a variety of different classes of heterocyclic compounds has been described to possess antagonistic activity at adenosine receptors, including xanthines, adenines, 7-deazaadenines, 7-deaza-8-azapurines, pyrazolo[3,4-*c*]quinolines, pyrazolo-[1,5-α]pyridines and 1,8-naphthyridines.[3–11] Isoquinoline and quinazoline derivatives from our laboratory also displayed high affinity for the adenosine ($A_1$ and $A_3$) receptors, in particular when a spacer-coupled aromatic group was attached to the core ring system.[12–14] With the recent synthesis of a series of (3-phenyl)-1,2,4-thiadiazoles, another new class of heterocyclic compounds as adenosine receptor antagonists was developed.[15] *N*-(3-Phenyl-1,2,4-thiadiazol-5-yl)-4-methoxybenzamide (**3**, LUF 5417) displayed similar adenosine $A_3$ receptor affinity as *N*-(3-phenylisoquinolin-1-yl)-4-methoxybenzamide (**2**, Fig. 2). However, compound **3** displayed a significant increase in affinity for the adenosine $A_1$ receptor, rendering **3** essentially non-selective. *N*-(4-Phenylthiazol-2-yl)-4-methoxybenzamide (**4**, LUF 5433, Fig. 2),[16] a single compound included in the study described,[15] showed a 9-fold selectivity for the adenosine $A_1$ receptor.

In the present study, we synthesised a series of 4-phenyl-(2-phenylcarboxamido)-1,3-thiazole derivatives as potential antagonists for the adenosine $A_1$ receptor on the basis of these findings. The synthesis of the substituted 4-phenyl-2-(phenylcarboxamido)-1,3-thiazoles **17**–**26** was achieved by a condensation reaction of 2-amino-4-phenyl-1,3-thiazole (**5**) with the appropriate acylchloride (**6**–**16**) yielding the corresponding amide-spaced compounds (Scheme 1). The HBr salt of 2-amino-4-phenyl-1,3-thiazole (193 mg, 0.7 mmol) was dissolved in dioxane (3 mL) and $Et_3N$ (195 μL). To this mixture the appropriate acylchloride (1.07 mmol) in 1 mL dioxane was added and the solution was refluxed overnight. After cooling to room temperature the mixure was



**1**

**Figure 1.**



**2** $K_i$ ($A_1$) 3000 ± 700 nM; $K_i$ ($A_3$) 79 ± 20 nM

**3** $K_i$ ($A_1$) 32 ± 5 nM; $K_i$ ($A_3$) 82 ± 4 nM

**4** $K_i$ ($A_1$) 76 ± 8 nM; $K_i$ ($A_3$) 670 ± 100 nM

**Figure 2.**

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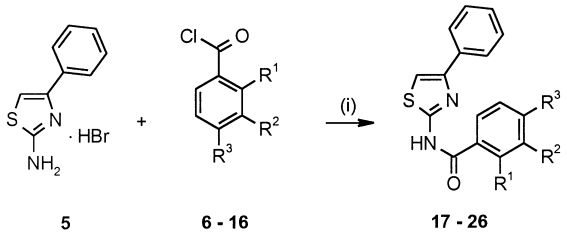
PII: S0960-894X(01)00356-0

filtered, concentrated under vacuo and purified by column chromatography. The target compounds were obtained in high yields.[17]

All compounds were tested in radioligand binding assays to determine their affinities for the adenosine $A_1$ receptor in rat brain cortex, the $A_{2A}$ receptor in rat striatum and the human $A_3$ receptor as expressed in HEK 293 cells (Table 1). Displacement experiments were performed in the absence of GTP. The procedures used have been described in detail previously.[18–20] Table 1 and binding data of the earlier mentioned series of thiadiazoles (**3**)[15] show that the 1,3-thiazole derivatives (**17**–**26**) have similar adenosine $A_1$ receptor affinities as their thiadiazole congeners. However, their $A_1$ receptor selectivity was increased significantly (Table 1). The unsubstituted 4-phenyl-2-(phenylcarboxamido)-1,3-thiazole (**17**)[16] showed affinities in the micromolar range at the adenosine $A_{2A}$ and $A_3$ receptor, while having high affinity for the $A_1$ receptor. Introduction of a methyl group (**22**)[16] did not influence adenosine receptor affinities, whereas a *tert*-butyl group (**23**) or a trifluoromethyl group (**24**) decreased the affinity for the adenosine $A_1$ receptor. The stronger electron-donating methoxy analogue (**4**) increased adenosine $A_{2A}$ and $A_3$ receptor affinity and thus has decreased adenosine $A_1$ receptor selectivity. Introduction of a halogen atom was more favorable at the *para*-position (**19**, **20**)[16] than at the *meta*-position (**18**)[21] for high adenosine $A_1$ receptor affinity. The 3,4-dichloro (**25**) and 2,4-dichloro (**26**) analogues showed affinities comparable to that of the unsubstituted compound (**17**). 2-[(4-Nitrophenyl)carboxamido]-4-phenyl-1,3-thiazole (**21**)[16] and 2-[(4-chlorophenyl)carboxamido]-4-phenyl-1,3-thiazole (**20**) had the highest affinity for the adenosine $A_1$ receptor ($K_i$ values of 22 and 18 nM, respectively). Their selectivity for adenosine $A_1$ versus $A_3$ receptors was approximately 50-fold.




**Scheme 1.**

**Table 1.** Affinities of 4-phenyl-2-(phenylcarboxamido)-1,3-thiazole derivatives (Scheme 1) at adenosine $A_1$, $A_{2A}$ and $A_3$ receptors expressed as $K_i$ values (nM ± SEM, $n = 3$) or percentage displacement

| | | | | $K_i$ (nM) or % displacement | | |
|---|---|---|---|---|---|---|
| No. | $R^1$ | $R^2$ | $R^3$ | $A_1$[a,e] | $A_{2A}$[b,e] | $A_3$[c,f] |
| **4** | H | H | $OCH_3$ | 76 ± 8[d] | 1900 ± 500[d] | 670 ± 100[d] |
| **17** | H | H | H | 39 ± 3 | 21% | 42% |
| **18** | H | Cl | H | 86 ± 1 | 3% | 42% |
| **19** | H | H | Br | 33 ± 4 | 14% | 58% |
| **20** | H | H | Cl | 18 ± 3 | 15% | 63% |
| **21** | H | H | $NO_2$ | 22 ± 4 | 9% | 35% |
| **22** | H | H | $CH_3$ | 36 ± 7 | 13% | 66% |
| **23** | H | H | $C(CH_3)_3$ | 1360 ± 80 | 2% | 27% |
| **24** | H | H | $CF_3$ | 165 ± 9 | 0% | 18% |
| **25** | H | Cl | Cl | 59 ± 8 | 6% | 20% |
| **26** | Cl | H | Cl | 58 ± 7 | 17% | 22% |

[a]Displacement of [$^3$H]DPCPX from rat cortical membranes.[20]
[b]Displacement of [$^3$H]ZM241385 from rat striatal membranes.[19]
[c]Displacement of [$^{125}$I]AB MECA from the human $A_3$ receptor expressed in HEK 293 cells.
[d]Data obtained from van Muijlwijk et al.[15]
[e]%Displacement at 10 μM.
[f]%Displacement at 1 μM.

Thus, in summary, the 4-phenyl-2-(phenylcarboxamido)-1,3-thiazole derivatives constitute a class of novel high affinity adenosine $A_1$ receptor antagonists. Furthermore, these compounds display higher $A_1$ selectivity compared to similar series previously described.

## Acknowledgements

The authors thank Karl-Norbert Klotz (University of Würzburg, Germany) for providing HEK 293 cells expressing the human adenosine $A_3$ receptor.

## References and Notes

1. Londos, C.; Cooper, D. M. F.; Wolff, J. *Biochemistry* **1980**, *77*, 2551.
2. Van Calker, D.; Müller, M.; Hamprecht, B. *J. Neurochem.* **1979**, *33*, 999.
3. Betti, L.; Biagi, G.; Giannaccini, G.; Giorgi, I.; Livi, O.; Lucacchini, A.; Manera, C.; Scartoni, V. *J. Med. Chem.* **1998**, *41*, 668.
4. Camaioni, E.; Costanzi, S.; Vittori, S.; Volpini, R.; Klotz, K.-N.; Cristalli, G. *Bioorg. Med. Chem.* **1998**, *6*, 523.
5. Collota, V.; Catarzi, D.; Varano, F.; Cecchi, L.; Filacchioni, G.; Martini, C.; Trincavelli, L.; Lucacchini, A. *J. Med. Chem.* **2000**, *43*, 3118.
6. Ferrarini, P. L.; Mori, C.; Manera, C.; Martinelli, A.; Mori, F.; Saccomanni, G.; Barili, P. L.; Betti, L.; Giannaccini, G.; Trincavelli, L.; Lucacchini, A. *J. Med. Chem.* **2000**, *43*, 2814.
7. Katsushima, T.; Nieves, L.; Wells, J. N. *J. Med. Chem.* **1990**, *33*, 1906.
8. Kuroda, S.; Akahane, A.; Itani, H.; Nishimura, S.; Durkin, K.; Kinoshita, T.; Tenda, Y.; Sakane, K. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 1979.
9. Müller, C. E. *Expert Opin. Ther. Pat.* **1997**, *5*, 419.
10. Pfister, J. R.; Belardinelli, L.; Lee, G.; Lum, R. T.; Milner, P.; Stanley, W. C.; Linden, J.; Baker, S. P.; Schreiner, G. *J. Med. Chem.* **1997**, *40*, 1773.
11. Poulsen, S.-A.; Quinn, R. J. *J. Med. Chem.* **1996**, *39*, 4156.
12. van Muijlwijk-Koezen, J. E.; Timmerman, H.; Link, R.; van der Goot, H.; IJzerman, A. P. *J. Med. Chem.* **1998**, *41*, 3987.
13. van Muijlwijk-Koezen, J. E.; Timmerman, H.; Link, R.; van der Goot, H.; IJzerman, A. P. *J. Med. Chem.* **1998**, *41*, 3994.
14. van Muijlwijk-Koezen, J. E.; Timmerman, H.; van der Goot, H.; Menge, W. M. P. B.; von Frijtag Drabbe Künzel, J.; de Groote, M.; IJzerman, A. P. *J. Med. Chem.* **2000**, *43*, 2227.
15. van Muijlwijk-Koezen, J. E.; Timmerman, H.; Vollinga, R. C.; von Frijtag Drabbe Künzel, J.; de Groote, M.; Visser, S.; IJzerman, A. P. *J. Med. Chem.* **2001**, *44*, 749.
16. Ramachandraiah, G.; Reddy, K.; Kuroda, S. *Indian J. Chem. Sect. B* **1985**, *24*, 808.

17. Physical data for the compounds: 2-[(4-Methoxyphenyl)carboxamido]-4-phenyl-1,3-thiazole (**4**): Yield 185 mg (0.60 mmol, 85%), mp 161–162 °C; $^1$H NMR (200 MHz, DMSO-$d_6$) δ 12.59 (bs, 1H, NH), 8.14 (d, 2H, $J$ = 8.92 Hz, $H_{ortho}$), 7.94 (d, 2H, $J$ = 7.56 Hz, $H_{meta}$), 7.65 (s, 1H, SCH), 7.47–7.32 (m, 3H, $H_{ortho+para}$), 7.08 (d, 2H, $J$ = 8.93 Hz, $H_{meta}$), 3.84 (s, 3H, $CH_3$) ppm; $m/z$ 311 ($M^+$); anal. ($C_{17}H_{14}N_2O_2S$) C.H.N. 4-Phenyl-2-(phenylcarboxamido)-1,3-thiazole (**17**): Yield 137 mg (0.49 mmol, 70%), mp 127 °C; $^1$H NMR (200 MHz, DMSO-$d_6$) δ 12.77 (bs, 1H, NH), 8.13 (d, 2H, $J$ = 7.90 Hz, $H_{arom}$), 7.95 (d, 2H, $J$ = 7.21 Hz, $H_{arom}$), 7.69 (s, 1H, SCH), 7.64–7.32 (m, 6H, $H_{arom}$) ppm; $m/z$ 281 ($M^+$); anal. ($C_{16}H_{12}N_2OS$) C.H.N. 2-[(3-Chlorophenyl)carboxamido]-4-phenyl-1,3-thiazole (**18**): Yield 176 mg (0.56 mmol, 80%), mp 136–137 °C; $^1$H NMR (200 MHz, DMSO-$d_6$) δ 12.89 (bs, 1H, NH), 8.19 (s, 1H, COCCH), 8.06 (d, 1H, $J$ = 7.89 Hz, CCHCCl), 7.96–7.92 (m, 2H, $H_{arom}$), 7.71 (s, 1H, SCH), 7.71–7.35 (m, 5H, $H_{arom}$) ppm; $m/z$ 315 ($M^+$); anal. ($C_{16}H_{11}ClN_2OS$) C.H.N. 2-[(4-Bromophenyl)carboxamido]-4-phenyl-1,3-thiazole (**19**): Yield 189 mg (0.53 mmol, 75%), mp 198–200 °C; $^1$H NMR (200 MHz, DMSO-$d_6$) δ 12.81 (bs, 1H, NH), 8.07 (m, 4H, $H_{arom}$), 7.78–7.69 (m, 3H, $H_{arom}$), 7.46–7.32 (m, 4H, $H_{arom}$) ppm; $m/z$ 360 ($M^+$); anal. ($C_{16}H_{11}BrN_2OS$) C.H.N. 2-[(4-Chlorophenyl)carboxamido]-4-phenyl-1,3-thiazole (**20**): Yield 183 mg (0.58 mmol, 83%), mp 202 °C; $^1$H NMR (200 MHz, DMSO-$d_6$) δ 12.85 (bs, 1H, NH), 8.13 (d, 2H, $J$ = 8.58 Hz, $H_{arom}$), 7.95 (d, 2H, $J$ = 7.21 Hz, $H_{arom}$), 7.70 (s, 1H, SCH), 7.63 (d, 2H, $J$ = 8.59 Hz, $H_{arom}$), 7.48–7.29 (m, 3H, $H_{arom}$) ppm; $m/z$ 315 ($M^+$); anal. ($C_{16}H_{11}ClN_2OS$) C.H.N. 2-[(4-Nitrophenyl)carboxamido]-4-phenyl-1,3-thiazole (**21**): Yield 180 mg (0.55 mmol, 79%), mp 213–214 °C; $^1$H NMR (200 MHz, DMSO-$d_6$) δ 13.02 (bs, 1H, NH), 8.39–8.26 (m, 4H, $H_{arom}$), 7.94 (d, 2H, $J$ = 8.24 Hz, $H_{ortho}$), 7.72 (s, 1H, SCH), 7.48–7.29 (m, 3H, $H_{meta+para}$) ppm; $m/z$ 326 ($M^+$); anal. ($C_{16}H_{11}N_3O_3S$) C.H.N. 2-[(4-Methylphenyl)carboxamido]-4-phenyl-1,3-thiazole (**22**): Yield 140 mg (0.48 mmol, 68%), mp 146–149 °C; $^1$H NMR (200 MHz, DMSO-$d_6$) δ 12.78 (bs, 1H, NH), 8.14 (d, 2H, $J$ = 7.89 Hz, $H_{ortho}$), 8.05 (d, 2H, $J$ = 7.89 Hz, $H_{ortho}$), 7.78 (s, 1H, SCH), 7.58–7.55 (m, 5H, $H_{arom}$), 2.59 (s, 3H, $CH_3$) ppm; $m/z$ 295 ($M^+$); anal. ($C_{17}H_{14}N_2OS$) C.H.N. 2-{[4-(*tert*-Butyl)phenyl]carboxamido}-4-phenyl-1,3-thiazole (**23**): Yield 167 mg (0.50 mmol, 71%); $^1$H NMR (200 Hz, DMSO-$d_6$) δ 12.69 (bs, 1H, NH), 8.08 (d, 2H, $J$ = 8.23 Hz, $H_{ortho}$), 7.95 (d, 2H, $J$ = 7.55 Hz, $H_{ortho}$), 7.66 (s, 1H, SCH), 7.57–7.28 (m, 5H, $H_{arom}$), 1.30 (s, 9H, $CH_3$) ppm; $m/z$ 337 ($M^+$); anal. ($C_{20}H_{20}N_2OS$) C.H.N. 4-Phenyl-2-{[4-(trifluoromethyl)phenyl]-carboxamido}-1,3-thiazole (**24**): Yield 146 mg (0.42 mmol, 60%), mp 198–200 °C; $^1$H NMR (200 MHz, DMSO-$d_6$) δ 13.04 (bs, 1H, NH), 8.38–8.22 (m, 2H, $H_{ortho}$), 8.05–7.80 (m, 3H, SCH, $H_{meta}$), 7.73–7.64 (m, 1H, $H_{para}$), 7.52–7.24 (m, 4H, $H_{arom}$) ppm; $m/z$ 349 ($M^+$); anal. ($C_{17}H_{11}F_3N_2OS$) C.H.N. 2-[(3,4-Dichlorophenyl)carboxamido]-4-phenyl-1,3-thiazole (**25**): Yield 164 mg (0.47 mmol, 67%), mp 192 °C; $^1$H NMR (200 MHz, DMSO-$d_6$) δ 12.89 (bs, 1H, NH), 8.42–8.34 (m, 1H, COCCHCH), 8.12–7.68 (m, 6H, SCH, $H_{arom}$), 7.50–7.25 (m, 3H, $H_{arom}$) ppm; $m/z$ 349 ($M^+$); anal. ($C_{16}H_{10}Cl_2N_2OS$) C.H.N. 2-[(2,4-Dichlorophenyl)carboxamido]-4-phenyl-1,3-thiazole (**26**): Yield 198 mg (0.57 mmol, 81%), mp 180–182 °C; $^1$H NMR (200 MHz, DMSO-$d_6$) δ 12.85 (bs, 1H, NH), 7.94–7.89 (m, 2H, $H_{arom}$), 7.79–7.69 (m, 3H, $H_{arom}$), 7.58–7.32 (m, 4H, $H_{arom}$) ppm; $m/z$ 349 ($M^+$); anal. ($C_{16}H_{10}Cl_2N_2OS$) C.H.N.

18. Briefly, assays for the human adenosine $A_3$ receptor were performed in 50/10/1 buffer [50 mM Tris/10 mM $MgCl_2$/1 mM ethylenediaminetetraacetic acid (EDTA) and 0.01% 3-([3-cholamidopropyl] - dimethylammonio) - 1 - propanesulfonate (CHAPS)] in glass tubes and contained 50 μL of a HEK 293 cell membrane suspension (10–30 μg), 25 μL [$^{125}$I]AB MECA (final concentration 0.15 nM), and 25 μL of ligand. Incubations were carried out for 1 h at 37 °C and were terminated by rapid filtration over Whatman GF/B filters, using a Brandell cell harvester (Brandell, Gaithersburg, MD, USA). Tubes were washed three times with 3 mL of buffer. Radioactivity was determined in a Beckman 5500B γ-counter. Nonspecific binding was determined in the presence of $10^{-5}$ M R-PIA.

19. Gao, Z.-G.; IJzerman, A. P. *Biochem. Pharmacol.* **2000**, *60*, 669.

20. Pirovano, I. M.; IJzerman, A. P.; Van Galen, P. J. M.; Soudijn, W. *Eur. J. Pharmacol.* **1989**, *172*, 185.

21. Coburn, R. A.; Glennon, R. A. *J. Pharm. Sci.* **1973**, *62*, 1785.